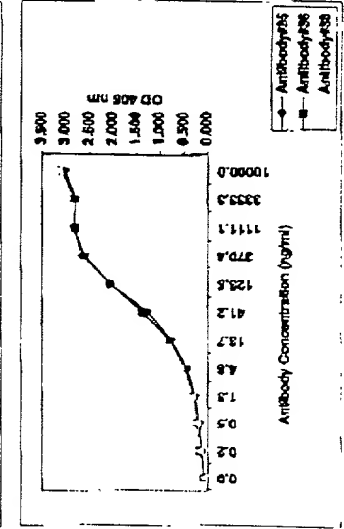
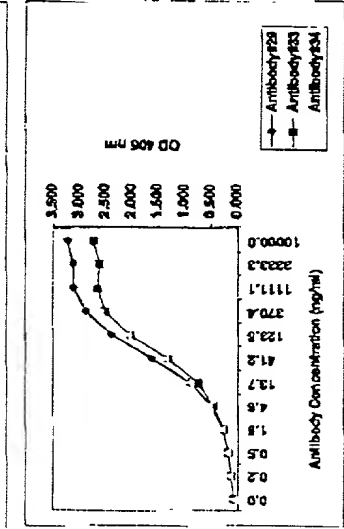
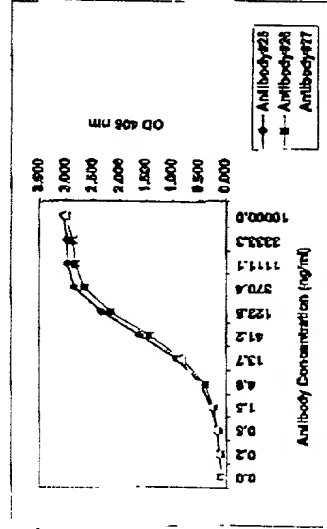
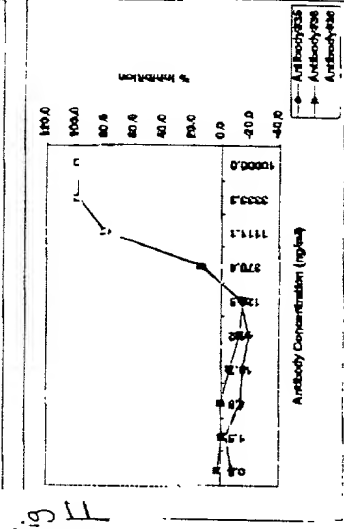
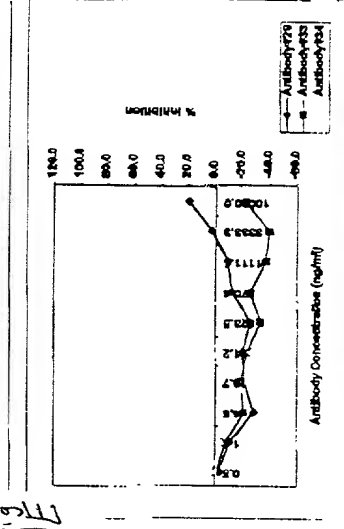
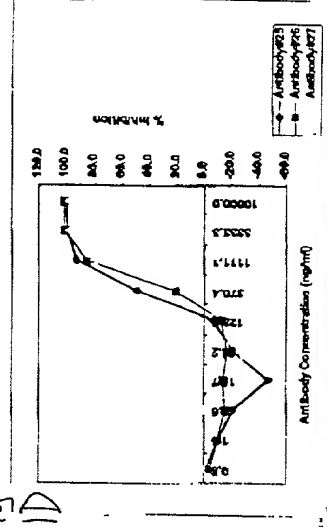


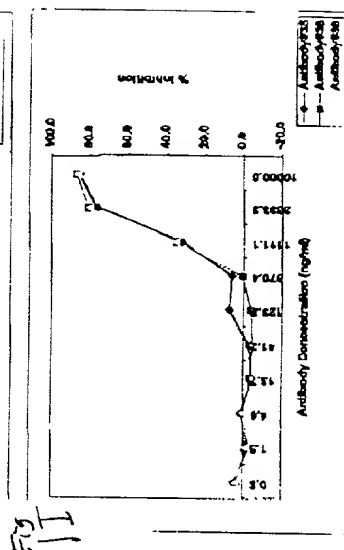
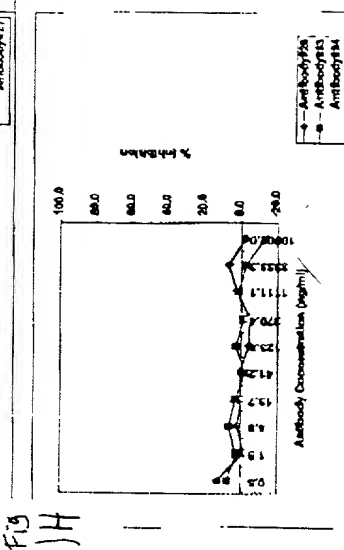
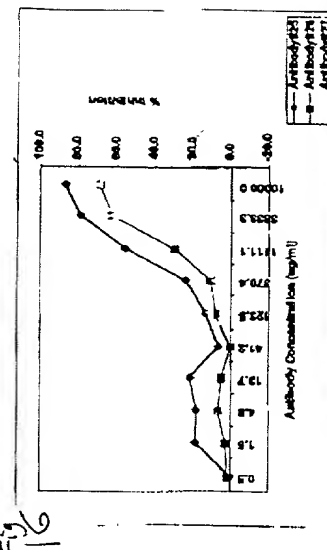
# Binding of antibodies to CTLA-4



# Blockage of CD86 binding



# Blockage of CD80 binding



Anti-CTLA4 Abs #25, #26, and #29 defined three distinct epitopes on CTLA4

CTLA4 point mutant	Antibody #25
Mock	0.165
WT	1.17
T67	0.935
Q82	1.1
R85	0.96
D63	0.988
R38	0.52
S64	1.07
I65	1.19
L61	1.14
T45	0.948
E46	0.811
D62	0.985
CTLA4-Ig	0.996
DME	0.14

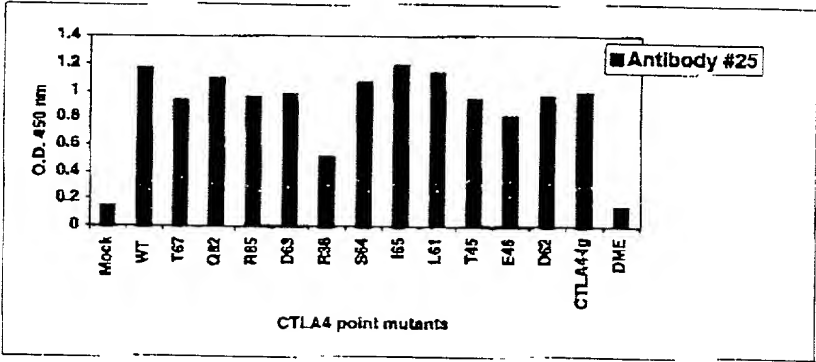


Fig.  
2A

CTLA4 point mutant	Antibody #26
Mock	0.174
WT	1.16
T67	0.944
Q82	1.07
R85	0.976
D63	0.979
R38	0.636
S64	1.1
I65	1.17
L61	1.19
T45	0.948
E46	0.18
D62	0.974
CTLA4-Ig	1.1
DME	0.163

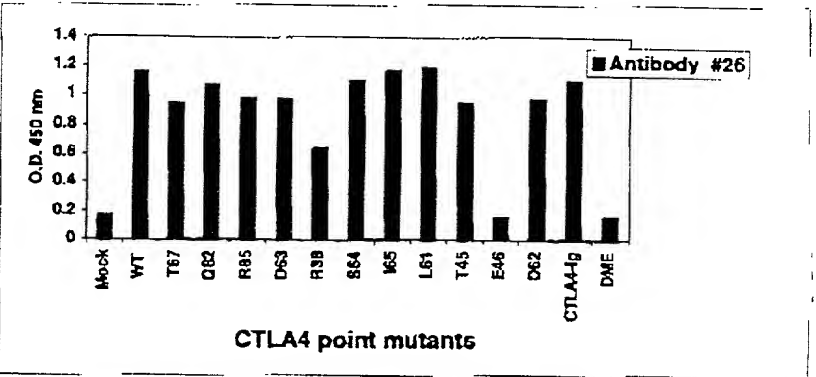


Fig.  
2B

CTLA4 point mutant	Antibody #29
Mock	0.094
WT	0.82
T67	0.695
Q82	0.784
R85	0.096
D63	0.719
R38	0.364
S64	0.753
I65	0.801
L61	0.819
T45	0.695
E46	0.609
D62	0.673
CTLA4-Ig	0.657
DME	0.091

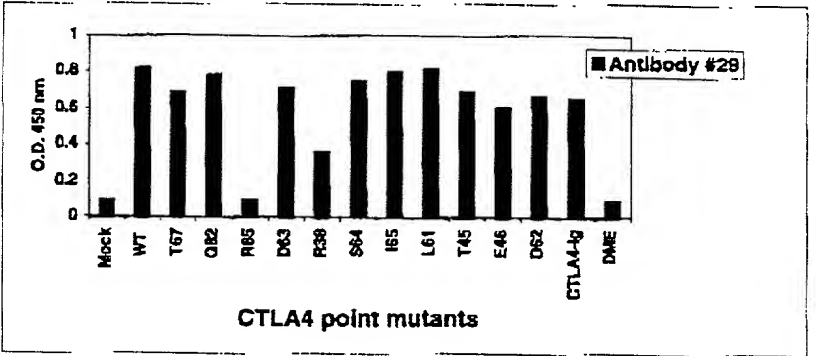


Fig.  
2C

Fig 3A Proliferation

	media	Ab #25	Ab #26	Ab#27	Ab#29	Ab#33	Ab#34	Ab#35	Ab#36	Ab #38
Donor 1	54871	16589	33928	49492	234	928	45524	56203	72358	50665
Donor 2	102159	32784.3	58736.2	79098.2	1891.3	5427.4	78391.9	91920.2	116454.6	88937.3

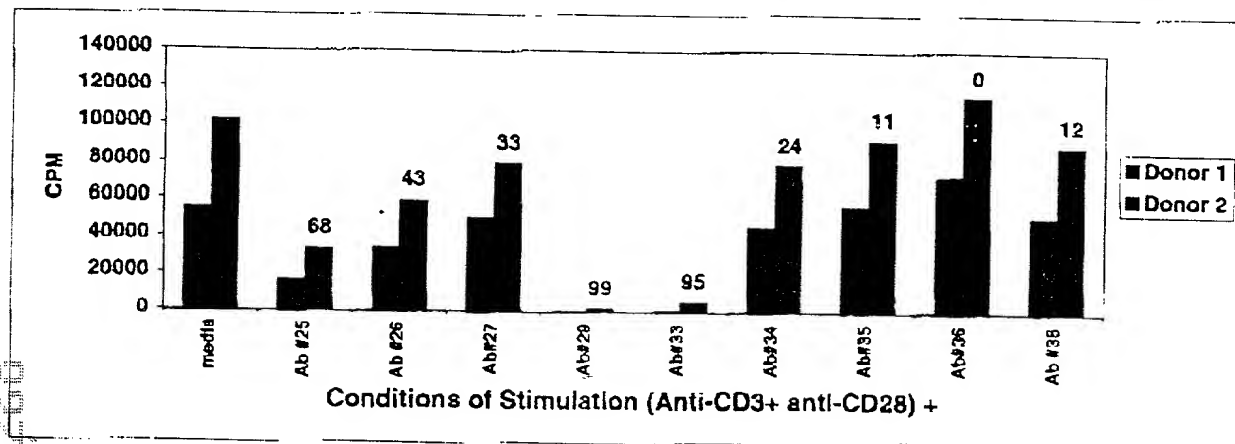


Fig 3B IL-2 Production

IL-2(pg/ml)	media	Ab #25	Ab #26	Ab#27	Ab#29	Ab#33	Ab#34	Ab#35	Ab#36	Ab #38
	2125.3	330.2	182.5	231.7	60.8	462.3	573.7	305.7	617.7	672.1

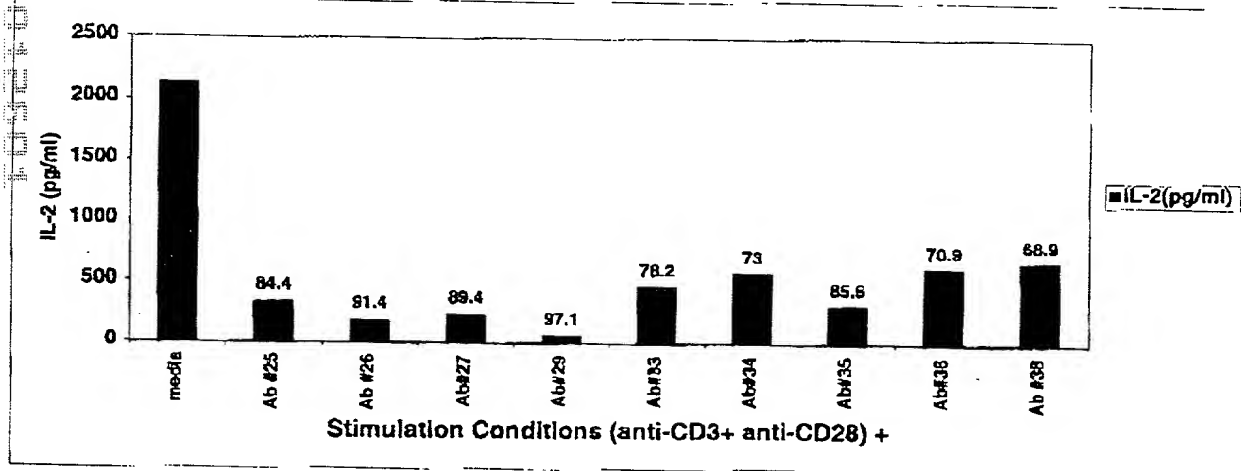


Fig. 4A Proliferation

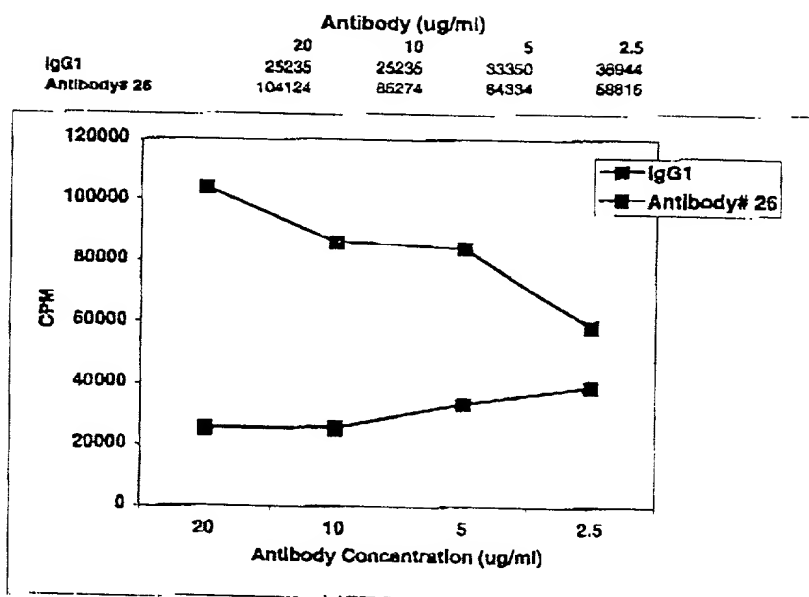
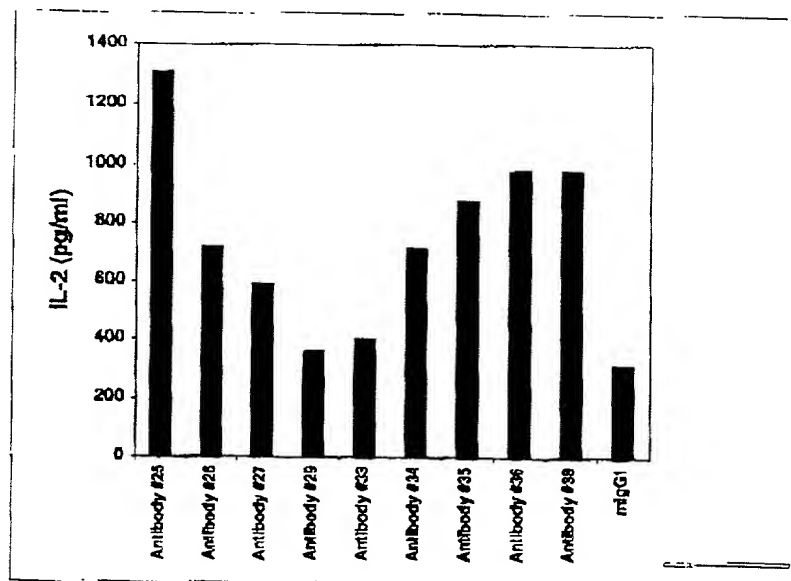


Fig 4B IL-2 Production

Antibody	IL-2 (pg/ml)
Antibody #25	1310
Antibody #26	720
Antibody #27	584
Antibody #29	358
Antibody #33	400
Antibody #34	715
Antibody #35	875
Antibody #36	982
Antibody #38	982
mgG1	317





FACS Competition Using FITC-Labeled CTLA4-26B with Cold CTLA4-26B and Humanized IgG1/κ

Name	Cell Line	Primary Ab	Competitor
052199.001	CHO.CTLA4	FITC-CTLA4-26B 125 ng	2000 ng
052199.002			1000 ng
052199.003			500 ng
052199.004			250 ng
052199.005			100 ng
052199.006	CHO.B7.1	none 125 ng none	IgG1/κ
052199.007			2000 ng
052199.008			1000 ng
052199.009			500 ng
052199.010			250 ng
052199.016			100 ng
052199.017			none
052199.018			none
052199.019			none

Fig 6A

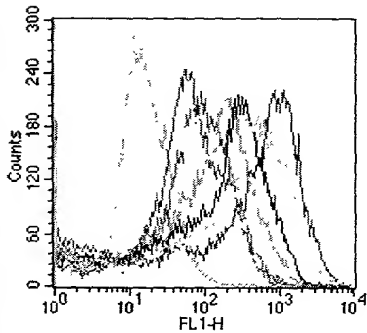


Fig 6B

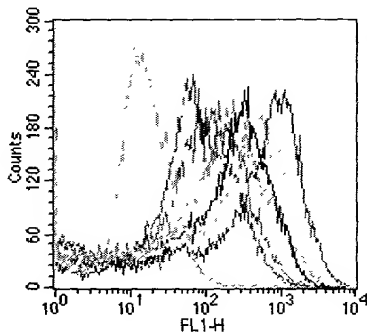
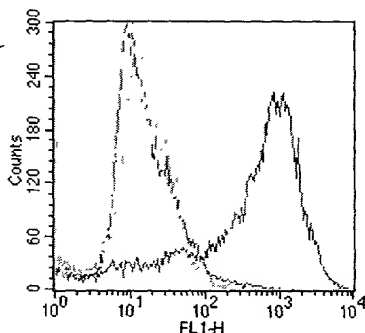


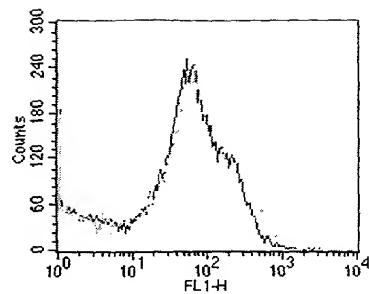
Fig 6C



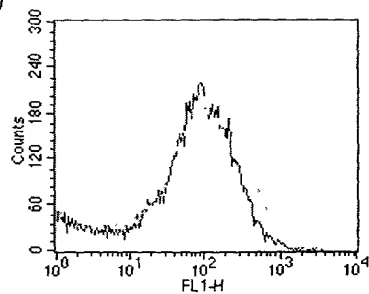
Key	Name	Parameter	Gate
---	052199.016	FL1-H	No Gate
---	052199.017	FL1-H	No Gate
---	052199.001	FL1-H	No Gate
---	052199.002	FL1-H	No Gate
---	052199.003	FL1-H	No Gate
---	052199.004	FL1-H	No Gate
---	052199.005	FL1-H	No Gate

Key	Name	Parameter	Gate
---	052199.016	FL1-H	No Gate
---	052199.017	FL1-H	No Gate
---	052199.006	FL1-H	No Gate
---	052199.007	FL1-H	No Gate
---	052199.008	FL1-H	No Gate
---	052199.009	FL1-H	No Gate
---	052199.010	FL1-H	No Gate

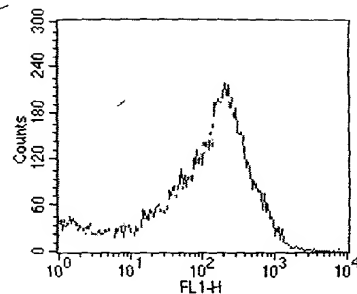
Key	Name	Parameter	Gate
---	052199.016	FL1-H	No Gate
---	052199.017	FL1-H	No Gate
---	052199.018	FL1-H	No Gate
---	052199.019	FL1-H	No Gate

Fig.  
7A

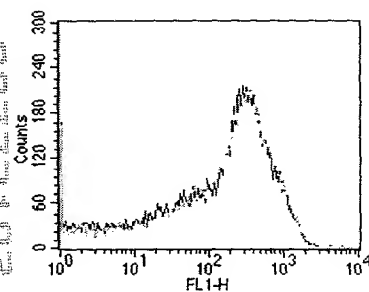
Key	Name	Parameter	Gate
—	052199.001	FL1-H	No Gate
—	052199.006	FL1-H	No Gate

Fig.  
7B

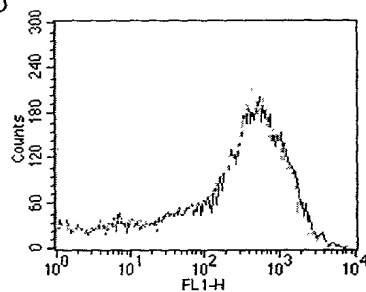
Key	Name	Parameter	Gate
—	052199.002	FL1-H	No Gate
—	052199.007	FL1-H	No Gate

Fig.  
7C

Key	Name	Parameter	Gate
—	052199.003	FL1-H	No Gate
—	052199.008	FL1-H	No Gate

Fig.  
7D

Key	Name	Parameter	Gate
—	052199.004	FL1-H	No Gate
—	052199.009	FL1-H	No Gate

Fig.  
7E

Key	Name	Parameter	Gate
—	052199.005	FL1-H	No Gate
—	052199.010	FL1-H	No Gate

Figure 7

Fig 8A

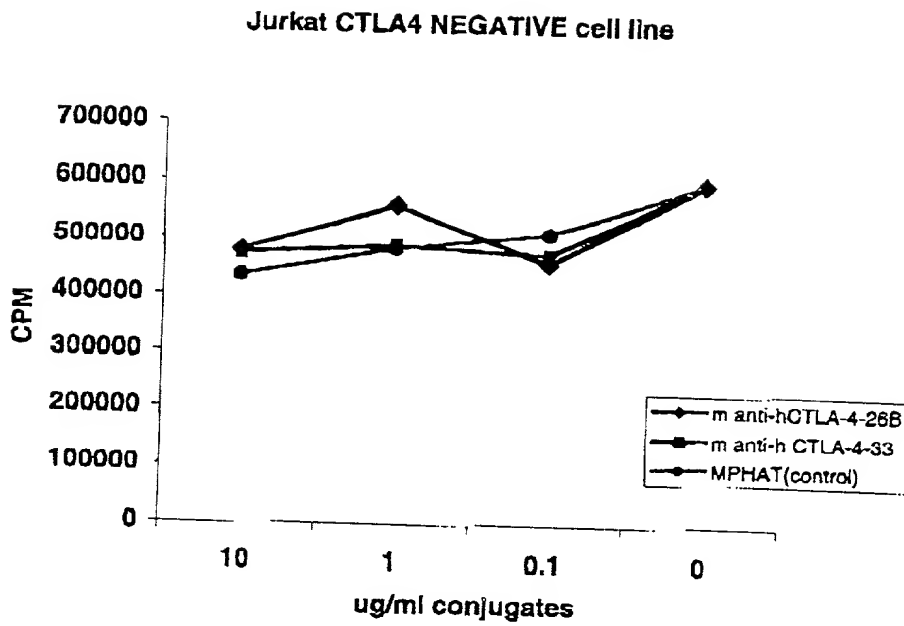
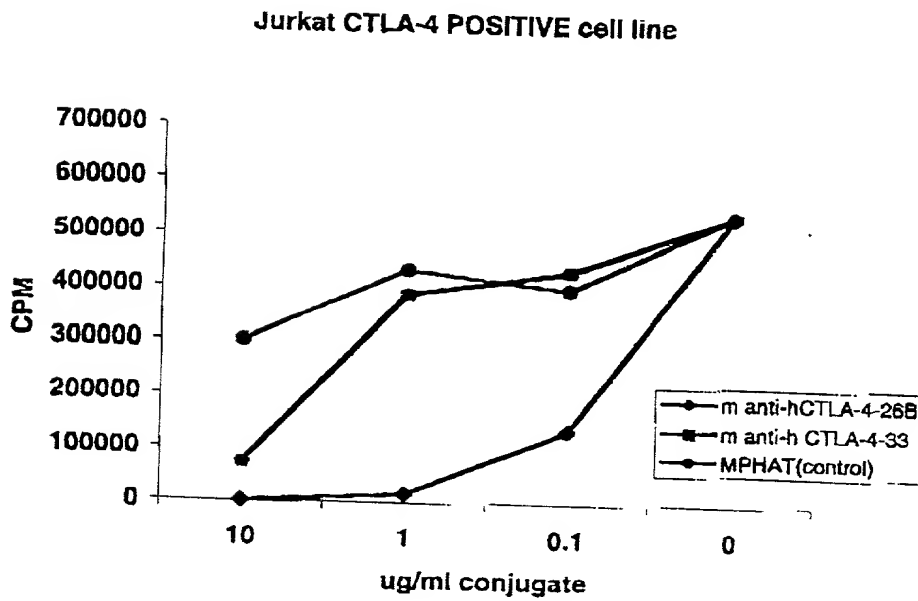


Fig 8B





HuCTLA4 - Vk

TATATCTAGACCACCATGGATTTTCAAGTGCAGATCTTCAGCTTCCTGCTAATCAGTGCC  
M D F Q V Q I F S F L L I S A

TCAGTCATACTGTCCAGAGGAGATATCCAGATGACCCAGTCTCCATCCTCCCTATCCGCA  
S V I L S R G D I Q M T Q S P S S L S A

TCTGTTGGGGACAGGGTCACCATAACCTGTAGTGCCACCTCAAGTATAACTTACATGTCC  
S V G D R V T I T C S A T S S I T Y M S

TGGTATCAGCAGAAGCCAGGAAAGGCTCCCAAGCTTCTGATTTATGACACATCCAACCTG  
W Y Q Q K P G K A P K L L I Y D T S N L

GCTTCTGGGGTACCTAGCCGCTTCAGTGGCAGTGGGTCTGGGACCGACTACACACTCACA  
A S G V P S R F S G S G S G T D Y T L T

ATCAGCAGCCTGCAGCCAGAAGATTTTGCCACTTATTACTGCCAGCAGTGGAGTAGTTAC  
I S S L Q P E D F A T Y Y C Q Q W S S Y

CCGCTCACGTTTCGGTGGAGGGACCAAGGTTGAGATAAAACGTAAGTAGAATCCAAAGTCT  
P L T F G G G T K V E I K

AGATATA

Figure 9

HuCTLA4 - VH

TATATCTAGACCACCATGGCTGTCCTGGTGCTGTTCTCTGCCTGGTGCATTTCCAAGC  
M A V L V L F L C L V A F P S

TGTGTCCTGTCCCAGGTGCAGCTGCAAGAGTCAGGACCTGGCCTGGTGAAGCCCTCACAG  
C V L S Q V Q L Q E S G P G L V K P S Q

ACACTGTCCTTGACTTGCACTGTCTCTGGGTTTTTCATTAACTCATATGGTGTATATTGG  
T L S L T C T V S G F S L T S Y G V Y W

GTTGCGCCAGCCTCCAGGAAAGGGTCTGGAGTGGCTGGGAGTAATATGGGCTGGTGGTACC  
V R Q P P G K G L E W L G V I W A G G T

ACAAATTATAATTCGGCTCTCATGTCCAGACTGACAATCAGCAAAGACACATCCAAGAAC  
T N Y N S A L M S R L T I S K D T S K N

CAAGTTTCCTTAAAACTCAGCAGTGTGACTGCAGCGGACACAGCCGTCTACTACTGTGCC  
Q V S L K L S S V T A A D T A V Y Y C A

CGAGGCCCCCGCACGCTATGATGAAGAGAGGCTATGCTATGGACTACTGGGGACAAGGA  
R G P P H A M M K R G Y A M D Y W G Q G

ACCCTAGTCACAGTCTCCTCAGGTGAGTCCTTAAACCTCTAGATATA  
T L V T V S S

Figure 10

